

Serial No. 09/936,173  
Amendment dated 28 November 2006  
Reply to Advisory Action mailed 26 September 2006  
and to Office Action mailed 12 April 2006

### *AMENDMENTS TO THE CLAIMS*

This listing of claims will replace all prior versions, and listings, of claims in the application.

#### ***Listing of Claims***

Claims 1-3 (canceled).

Claim 4 (currently amended): The method of claim 31 wherein the concentration of the ~~multi-effect triazole~~ MET is about 0.1 mg/l.

Claim 5 (currently amended): The method of claim 31 wherein the ~~cotton seedlings of~~ medium in step (a) are grown in medium further comprising comprises about 0.01 to 0.2 mg/l of  $\alpha$  naphthalene acetic acid (NAA).

Claim 6 (canceled).

Claim 7 (currently amended): The method of claim 6 5 wherein the concentration of  ~~$\alpha$  naphthalene acetic acid~~ NAA is 0.05 mg/l.

Claim 8 (currently amended): The method of claim 31 wherein ~~the step (e) of regenerating the somatic embryos into whole plants~~ is carried out in the presence of about 0.05 to 0.2 mg/l of ~~multi-effect triazole~~ MET.

Claim 9 (canceled).

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Claim 10 (previously presented): The method of claim 8 wherein the concentration of the ~~multi-effect triazole~~ MET is 0.1 mg/l.

Claim 11 (currently amended): The method of claim 8 wherein ~~the step (e) of regenerating the somatic embryos into whole plants~~ is further carried out in the presence of about 0.01 to 0.2 mg/l  $\alpha$  naphthalene acetic acid (NAA).

Claim 12 (canceled).

Claim 13 (previously presented): The method of claim 11 wherein the concentration of  $\alpha$  naphthalene acetic acid NAA is 0.05 mg/l.

Claim 14 (currently amended): The method of claim 31 wherein ~~the step (b) of inducing callus formation~~ is carried out in a callus inducing culture medium comprising myo-inositol, vitamin B<sub>1</sub> and ~~a dimethylallyl (amino) purine~~ 2iP.

Claim 15 (currently amended): The method of claim 31 wherein ~~the step (d) of inducing somatic embryo formation~~ is carried out in a somatic embryo inducing culture medium comprising myo-inositol, vitamin B<sub>1</sub> and ~~a dimethylallyl (amino) purine~~ 2iP.

Claim 16 (currently amended): The method of claim 14 wherein the callus inducing culture medium comprises from about 50 to 150 mg/L of myo-inositol, from about 0.2 to 10 mg/L vitamin B<sub>1</sub> and from about 0.1 to 7.5 mg/L ~~dimethylallyl (amino) purine~~ 2iP.

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Claim 17 (currently amended): The method of claim 16 wherein the callus inducing culture medium comprises 100 mg/L myo-inositol, 0.4 mg/L vitamin B<sub>1</sub> and 5 mg/L ~~dimethylallyl (amino)~~ purine 2iP.

Claim 18 (currently amended): The method of claim 15 wherein somatic embryo inducing culture medium comprises from about 50 to 100 mg/L myo-inositol, from about 0.2 to 10 mg/L vitamin B<sub>1</sub> and from about 0.1 to 0.5 mg/L ~~dimethylallyl (amino)~~ purine 2iP.

Claim 19 (currently amended): The method of claim 18 wherein somatic embryo inducing culture medium comprises 100 mg/L myo-inositol, 0.4 mg/L vitamin B<sub>1</sub> and 5 mg/L ~~dimethylallyl (amino)~~ purine 2iP.

Claim 20 (currently amended): The method of claim 31 wherein ~~the step (b) of inducing callus formation~~ is carried out in a callus inducing culture medium comprising vitamin B<sub>5</sub>, ~~(2,4-dichlorophenoxy) acetic acid~~ 2,4-D, MgCl<sub>2</sub> and glucose.

Claim 21 (currently amended): The method of claim 31 wherein ~~the step (d) of inducing somatic embryo formation~~ is carried out in a somatic embryo inducing culture medium comprising vitamin B<sub>5</sub>, ~~(2,4-dichlorophenoxy) acetic acid~~ 2,4-D, MgCl<sub>2</sub> and glucose.

Claim 22 (currently amended): The method of claim 20 wherein the callus inducing culture medium comprises from about 0.2 to 10 mg/L vitamin B<sub>5</sub>, from about 0.05 to 0.15 mg/L ~~(2,4-dichlorophenoxy) acetic acid~~ 2,4-D, from about 0.4 to 1.2 mg/L, MgCl<sub>2</sub> from about 1% to 5% glucose.

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Claim 23 (currently amended): The method of claim 22 wherein the callus inducing culture medium comprises 0.4 mg/L vitamin B<sub>5</sub>, 0.1 mg/L (~~2,4-dichlorophenoxy~~) acetic acid 2,4-D, 0.8 mg/L MgCl<sub>2</sub> and 3% glucose.

Claim 24 (currently amended): The method of claim 21 wherein the somatic embryo inducing culture medium comprises from about 0.2 to 10 mg/L vitamin B<sub>5</sub>, from about 0.05 mg/L to 0.15 mg/L (~~2,4-dichlorophenoxy~~) acetic acid 2,4-D, from about 0.4 to 1.2 mg/L, MgCl<sub>2</sub> from about 1% to 5% glucose.

Claim 25 (currently amended): The method of claim 24 wherein the somatic embryo inducing medium comprises 0.4 mg/L vitamin B<sub>5</sub>, 0.1 mg/L (~~2,4-dichlorophenoxy~~) acetic acid 2,4-D, 0.8 mg/L MgCl<sub>2</sub> and 3% glucose.

Claim 26 (previously presented): A method according to claim 31, wherein the medium of steps (a), (b), (c), (d) or (e) further comprises from about 1.0 g/L to 3.0 g/L gellan gum.

Claim 27 (canceled).

Claim 28 (currently amended): The method of claim 31 wherein the step of inducing somatic embryo culture is carried out in a somatic embryo-inducing medium comprising a nitrate in an amount from about 1900 to ~~3800~~ 5700 mg/L.

Claim 29 (canceled).

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Claim 30 (previously presented): A method according to claim 28, wherein the nitrate is  $\text{KNO}_3$ .

Claim 31 (currently amended): A method for producing a transgenic cotton plant comprising:

(a) preparing explants from fibrous roots of cotton seedlings cultured in medium comprising about 0.05 mg/l to 0.2 mg/l of multi-effect triazole (MET);

(b) culturing said root explants in medium comprising a plant hormone selected from the group consisting of (i) 2,4-dichlorophenoxy (2,4-dichlorophenoxy) acetic acid (2,4-D), (ii) 6- $\gamma\gamma$ -dimethylallyl(amino) purine (2iP), (iii) a mixture of 2,4-D and kinetin and (iv) a mixture of 2iP and  $\alpha$  naphthalene acetic acid to induce callus formation;

(c) transforming said callus with *Agrobacterium tumefaciens* comprising a first DNA encoding a chimeric gene of interest to effect the stable transfer of said chimeric gene to the genome of cells comprising the callus tissue;

(d) culturing said transformed callus to induce inducing somatic embryos from said transformed callus and development of plantlets from said somatic embryos; and

(e) regenerating whole rooting said plantlets to produce transgenic cotton plants having said gene of interest from said somatic embryos.

Claim 32 (currently amended): The method of claim 31 wherein said DNA ~~encodes~~ is selected from the group consisting of an herbicide resistance gene, a gene that confers glyphosate resistance, a shikimate synthase gene and a *Bacillus thuringiensis* toxin gene.

Claims 33-35 (canceled).

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Claim 36 (currently amended): The method of claim 31 wherein ~~callus derived from explants of cotton seedling fibrous roots is transformed with said~~ *Agrobacterium tumifaciens* ~~comprising a first DNA encoding a chimeric gene of interest and further comprises~~ a second DNA encoding a selectable marker gene to effect the stable transfer of ~~said chimeric gene and~~ said selectable marker gene to the genome of cells comprising the callus tissue.

Claim 37 (new): The method of claim 31, wherein said seedlings are seedlings of *Gossypium hirsutum* cv. Coker 312.

Claim 38 (new): The method of claim 28, wherein the amount of nitrate is about 3800 mg/L.

Claim 39 (new): A method according to claim 38, wherein the nitrate is  $\text{KNO}_3$ .